***Table 1.*** Strains and plasmids used.

| Strain / plasmids | Description | Reference / source |
|---|---|---|
| *S. coelicolor* | | |
| M145 | Parental strain SCP1$^-$ SCP2$^-$ | Hopwood *et al.* (1985) |
| T124 | M145 (*accB*:pTR124), Th$^R$, Hyg$^R$ | This work |
| T149 | T124 containing pTR149 integrated in the *att* site of $\phi$C31, Th$^R$, Hyg$^R$, Am$^R$ | This work |
| T149A | T149 with the wilde type *accB* copy of the chromosome replaced by the *accB*::*hyg* mutant allele, Hyg$^R$, Am$^R$ | This work |
| *E. coli* | | |
| DH5$\alpha$ | F$^-$ $\Delta$*lacU169* ($\phi$80*lacZ*$\Delta$M15) *endA1 recA1 hsdR17 deoR supE44 thi-1* $\lambda^-$ *gyrA96 relA1* | Hanahan (1983) |
| BL21$\lambda$(DE3) | F$^-$ *ompT* $r_B^-$ $m_B^-$ (DE3) | Studier & Moffatt (1986) |
| ET 12567 | *supE44 hsdS20* ($r^-_Bm^-_B$) *ara-14 pro A2 lacY galK2 rpsL20 xyl-5 mtl-1 dam*$^-$ *dcm*$^-$ *hsdM*$^-$ Cm$^R$ | MacNeil *et al.* (1992) |
| RG7 | DH5$\alpha$ carrying pCL1 and pBA11 plasmids | Rodriguez & Gramajo (1999) |
| Plasmids | | |
| pBluescript SK(+) | Phagemid vector (Ap$^R$ *lacZ'*) | Stratagene |
| pGEM-T Easy | For cloning PCR products | Promega |
| pIJ2925 | pUC18 derivative (Ap$^R$ *lacZ'*) | Janssen & Bibb (1993) |
| pSET151 | For the conjugal transfer of DNA from *E. coli* to *Streptomyces* spp. (Ap$^R$ Th$^R$ *lacZ'*) | Bierman *et al.* (1992) |
| pET22b(+) | Phagemid vector (Ap$^R$ *lacZ'*) for expression of recombinant proteins under control of strong T7 transcription and translation signals | Novagen |
| pUZ8002 | RK2 derivative with defective *oriT* (Km$^R$) | Paget *et al.* (1999) |
| pIJ8600 | For the conjugal transfer of DNA from E. coli to Streptomyces spp. and for expression of recombinant proteins under *tipA* promoter | Sun *et al.* (1999) |
| pBA11 | Vector containing *E. coli birA* gene | Barker & Campbell (1981) |
| pCL1 | pSK(+) with a *Eco*RI-*Kpn*I insert carrying *accA1* | Rodriguez & Gramajo (1999) |
| pMR08 | pSK(+) with a *Sst*I insert carrying *accBE* | This work |
| pTR88 | pET22b(+) with *accBE* under control of strong T7 transcription and translation signals | This work |
| pTR90 | pET22b(+) with *accB* under control of strong T7 transcription and translation signals | This work |
| pTR107 | pET22b(+) with *accE* under control of strong T7 transcription and translation signals | This work |
| pTR124 | pSET151 with a *hyg* (Hyg$^R$) gene inserted in the *accB* coding region | This work |

| | | |
|---|---|---|
| pTR141 | pIJ8600 derivative carrying *oriT* RK2, *ori* pUC18, *attP* site, *int* ϕC31 and *aac(3)IV* ($Am^R$) | This work |
| pTR149 | pTR141 with a *KpnI* insert carrying *accBE* | This work |

**Table 2.** Heterologous expresion of acyl-CoA carboxylase components in cell-free extracts of *E. coli* and *in vitro* reconstitution of enzyme activity

| Strain | Proteins | Cell-free extracts | |
|---|---|---|---|
| *E. coli* * | induced by IPTG | ACCase [mU (mg protein) $^{-1}$]$^+$ | PCCase [mU (mg protein)$^{-1}$]$^+$ |
| RG7 | AccA1+BirA | ND | ND |
| RG8 | AccB, AccE | ND | ND |
| RG9 | AccB | ND | ND |
| RG10 | AccE | ND | ND |
| RG7:RG8 & | AccA1+BirA:AccB, AccE | 2.35±0.06 | 3.10±0.07 |
| RG7:RG9 & | AccA1+BirA:AccB | 0.32±0.05 | 0.50±0.05 |
| RG7:RG9:RG10 & | AccA1+BirA:AccB:AccE | 1.38±0.05 | 1.77±0.06 |

ND, Not detectable. The amount of $^{14}C$ fixed into acid-stable products was not significantly higher than background levels (10-30 c.p.m., equivalent to 0.02-0.06 mU).

* All the RG strains are derived from *E. coli* DH5α

+ Results are means of three determinations ± SE

++ pBA11 expresses BirA constitutively

& Mix of equal amount of proteins from cell-free extracts from each of the strains indicated

**Table 3 ACCase and PCCase activities in M145, M86 and M94**

| Strain *S. coelicolor* | Induction with Thiostrepton | Activity | |
|---|---|---|---|
| | | ACCase [mU (mg protein)$^{-1}$]* | PCCase [mU (mg protein)$^{-1}$]* |
| M145 | - | 1.12±0.03 | 2.2±0.03 |
| M86 | - | 0.43±0.03 | 1.45±0.06 |
| M86 | + | 0.33±0.03 | 0.95±0.06 |
| M94 | - | 0.40±0.03 | 1.57±0.03 |
| M94 | + | 4.61±0.03 (11.5) | 5.41±0.03 (3.5) |

* Results are means of three determinations ± SE.

**Table 4 Production of actinorhodin and undecylprodigiosin in YEME medium by M145 and M94.**

| Time (h) | M145 | | M94 | | [M94 +1µg Th]* | | [M94 + 5 µg Th]* | |
|---|---|---|---|---|---|---|---|---|
| | Act | Red | Act | Red | Act (M) | Red (M) | Act (M) | Red (M) |
| 40 | - | - | - | - | $3x10^{-7}$ | $10x10^{-7}$ | $11x10^{-7}$ | $28x10^{-7}$ |
| 60 | - | - | - | - | $9x10^{-7}$ | $12x10^{-7}$ | $26x10^{-7}$ | $78x10^{-7}$ |

* Results are mean of two indepent determinations.

(-) Means no detection of the antibiotics.

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